

Evidence on the Developmental and Reproductive Toxicity of Sulfur Dioxide

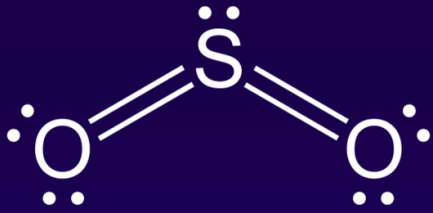
July 12, 2011

Developmental and Reproductive Toxicant
Identification Committee (DART) Meeting



Reproductive and Cancer Hazard Assessment Branch
Office of Environmental Health Hazard Assessment
California Environmental Protection Agency





Sulfur Dioxide (SO₂)

- Colorless, nonflammable gas, pungent odor
- In air pollution – in combination with sulfuric acid, sulfur trioxide, ozone, nitrogen dioxide, and particulates
- In ambient air from
 - fossil fuel consumption at power generation and other industrial facilities
 - wildfires

Sulfur Dioxide (SO₂)

- Criteria air pollutant
- US EPA SO₂ standards
 - New standard
 - 1-hour period: 0.075 parts per million (75 ppb)
- Primary route of exposure
 - Inhalation of gaseous SO₂
 - Smaller percentage absorbed at low (~40% at 0.001 ppb) vs high air concentrations (>90% at 0.100 ppb)

Male Reproductive Toxicity

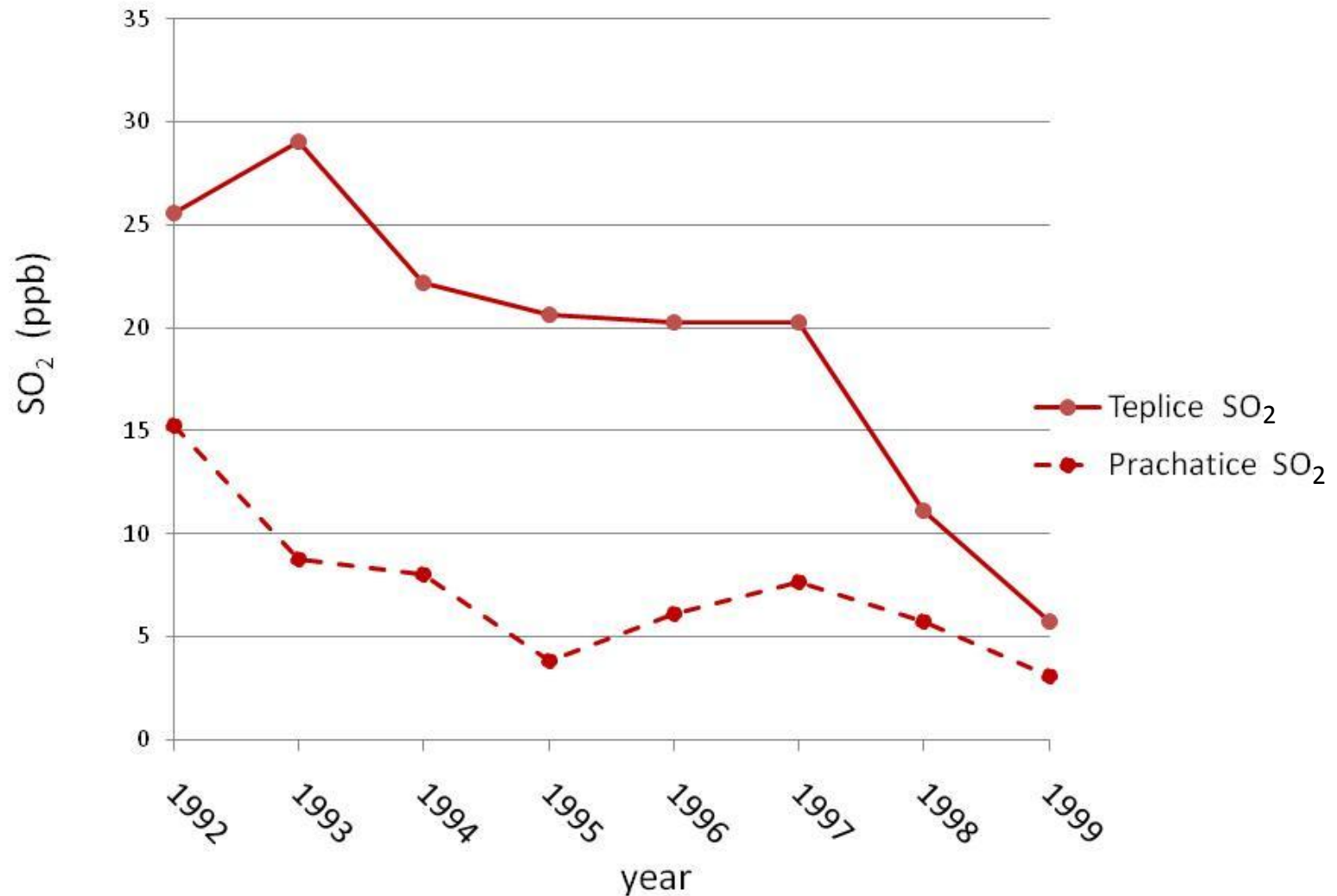
Human Studies

Teplice Program (1991-1999)

- Czech government / U.S. EPA
- Very high levels of air pollution



SO₂ Levels in Teplice and Prachatice



Benes et al., 2001

Male Reproductive Toxicity Studies in Humans

Retrospective cohort study

- fecundability

- Studies on sperm

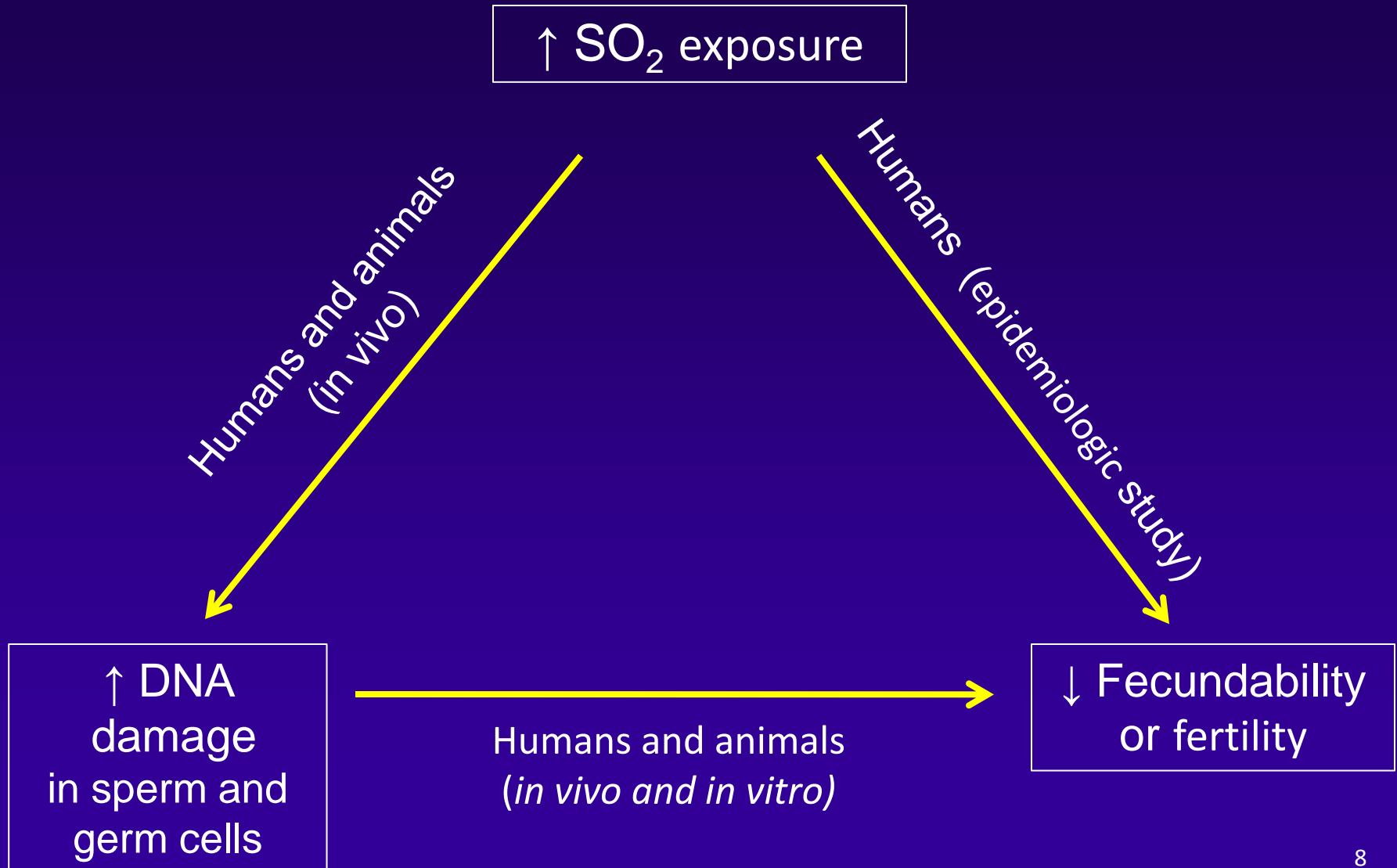
- quality

- genetic integrity

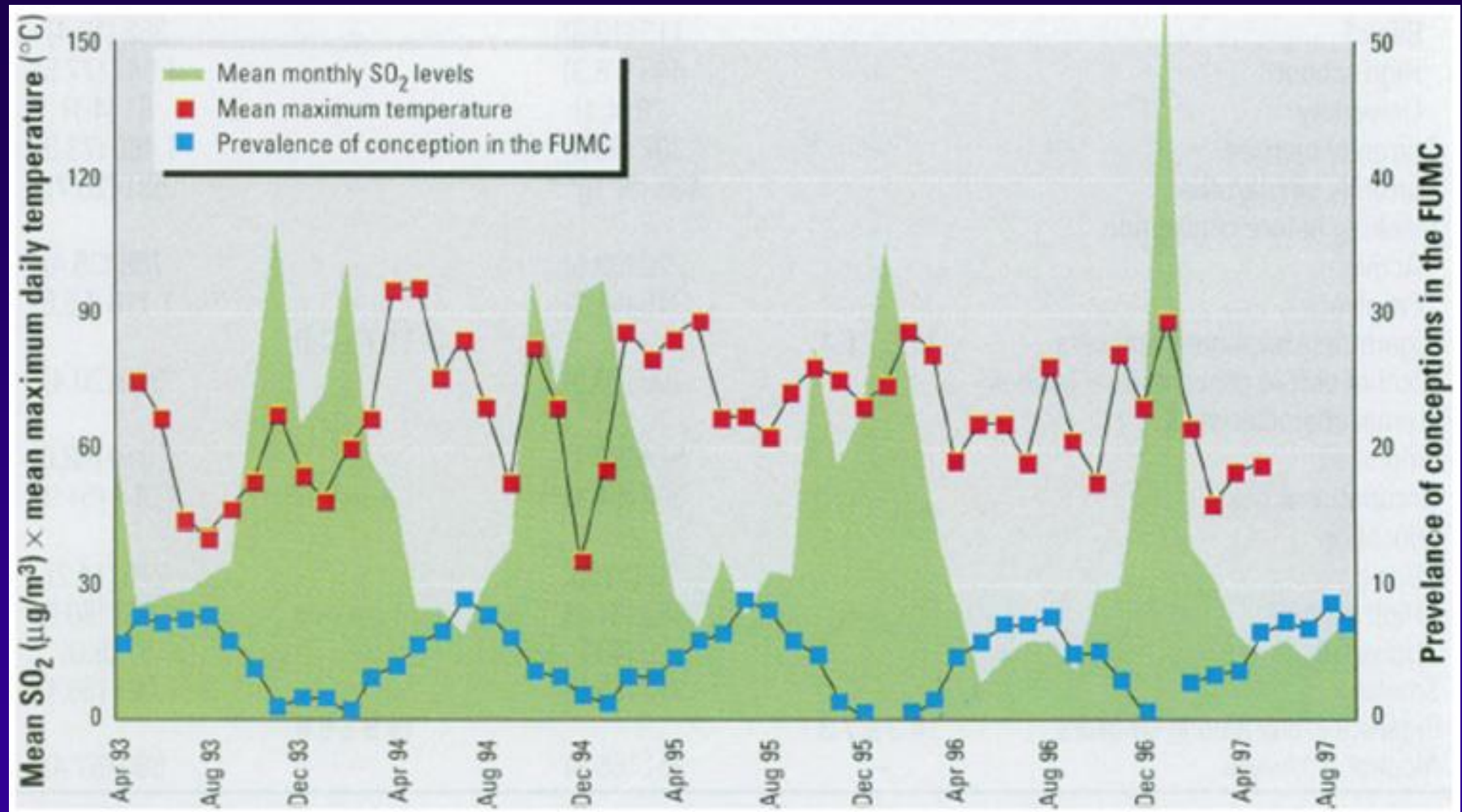
- DNA – abnormal chromatin structure

- aneuploidy

Male Reproductive Toxicity

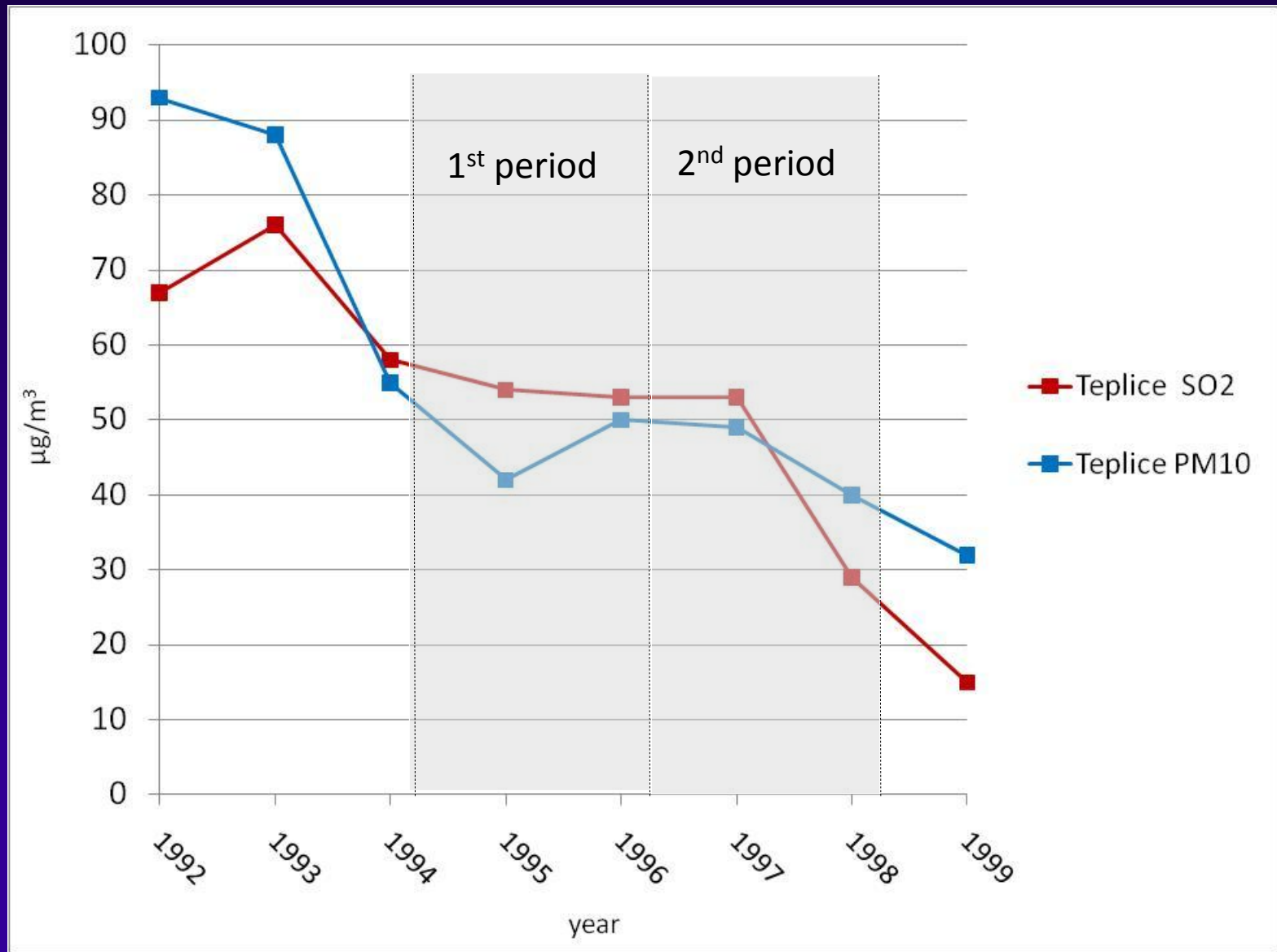


SO₂ levels, Temperature, and Conception



30-day running averages of SO₂ levels (in micrograms per cubic meter) and 30-day maximal daily temperatures (°C) compared to percent conception in the FUMC by the second month before conception

SO₂ Levels in Teplice



Adjusted odds ratios (AOR) for conceiving in the first unprotected menstrual cycle by exposure to SO₂ prior to conception

	Medium 15.3 – 30.5 ppb			High > 30.5 ppb		
	AOR	CI	p-value	AOR	CI	p-value
1st 2-yr period						
4 mos	1.58	0.85–2.74	0.16	1.26	0.58–2.71	0.56
3 mos	0.88	0.49–1.57	0.66	0.86	0.41–1.82	0.70
2 mos	0.49	0.25–0.96	0.037	0.43	0.20–0.93	0.033
1 mos	1.14	0.67–1.97	0.62	1.20	0.58–2.48	0.62
2nd 2-yr period						
4 mos	0.90	0.51–1.61	0.74	0.88	0.41–1.85	0.73
3 mos	0.85	0.45–1.57	0.59	0.96	0.45–2.03	0.91
2 mos	0.67	0.36–1.28	0.22	0.59	0.36–1.28	0.20
1 mos	1.16	0.59–2.29	0.66	1.15	0.59–3.59	0.31

Influence of distance from the monitor on the adjusted odds ratios of conceiving in the second month (30-60 days) before conception

Distance	Medium			High		
	AOR	CI	p-value	AOR	CI	p-value
< 3.5km	0.56	0.31–1.00	0.05	0.36	0.17–0.73	0.005
> 3.5km	0.58	0.31–1.08	0.09	0.70	0.34–1.45	0.34

Evidence of a Causal Association

- Reduced odds of conception with SO₂ exposure >15.3 ppb in 2nd month before conception
- Timing of the effect coincides with critical period
- Dose-response association with SO₂ exposure
- Stronger association when considering distance
- Decreased fecundability only seen with SO₂
- Effects on sperm motility and morphology appeared reversible

Human Studies on Sperm

- Air pollution - adverse effects on sperm quality and sperm chromatin
- SO₂ increased DNA damage in sperm
- SO₂ increased risk of aneuploidy in sperm

Animal Studies Male Reproductive Toxicity

Mice exposed to SO₂ by inhalation showed toxic effects in the testis, as well as other organs:

- Altered testis basement membranes, damaged Sertoli cells and spermatids ($\geq \sim 11,000$ ppb)
- Altered testicular biochemical parameters ($\geq \sim 8,400$ ppb), increased DNA damage
- ↑ levels of lipid peroxidation, altered intracellular redox status in mouse organs, including testes (at $\sim 20,000$ ppb)

Summary

Male Reproductive Toxicity

- Decreased fecundability
- Decreased sperm quality
- Toxic effects to the testes
- Increased DNA damage in sperm

Questions?

Developmental Toxicity

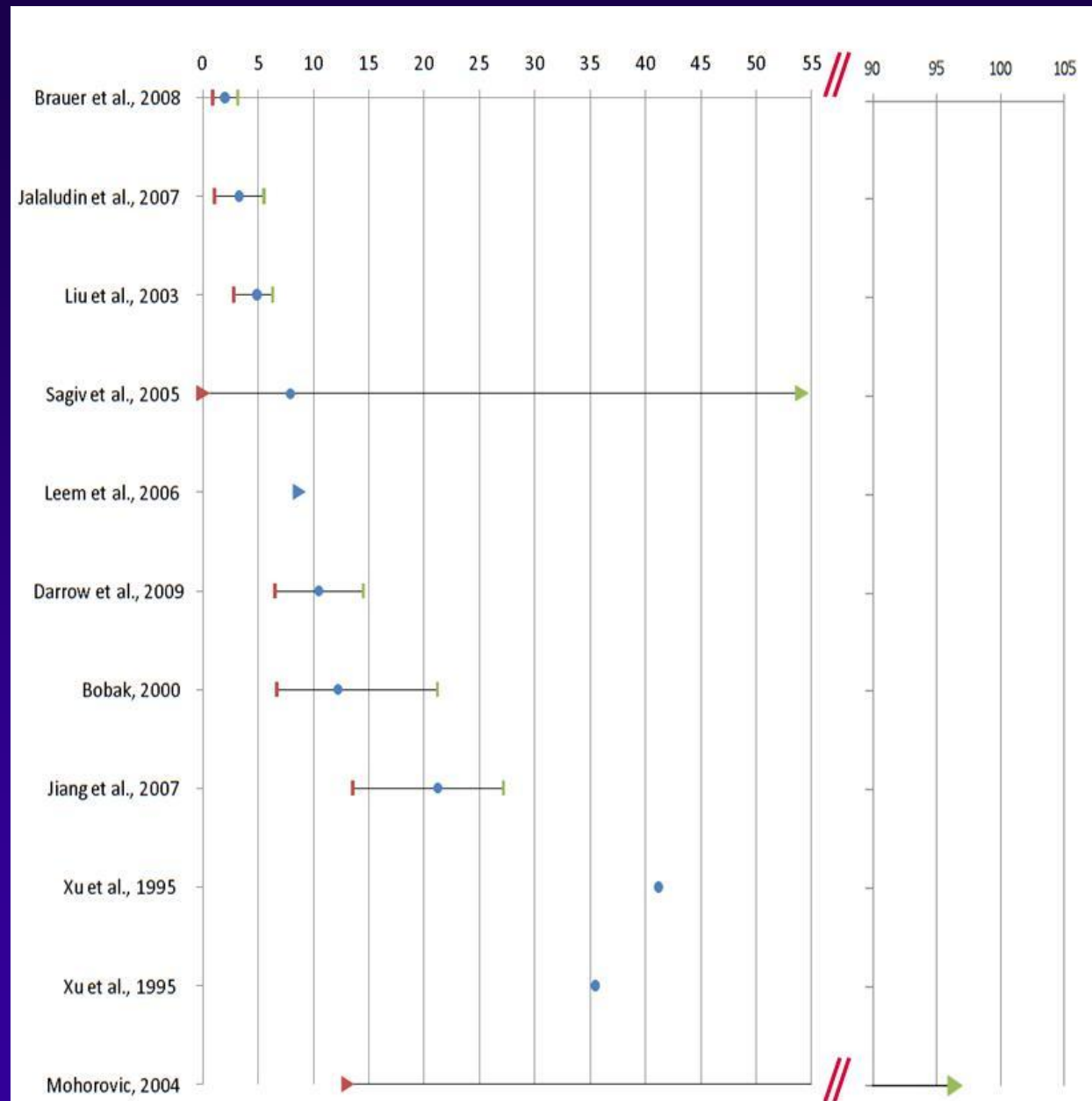
Developmental Toxicity

- Preterm birth
- Low birth weight
- Congenital malformations
- Pregnancy loss
- Asthma
- Other developmental effects

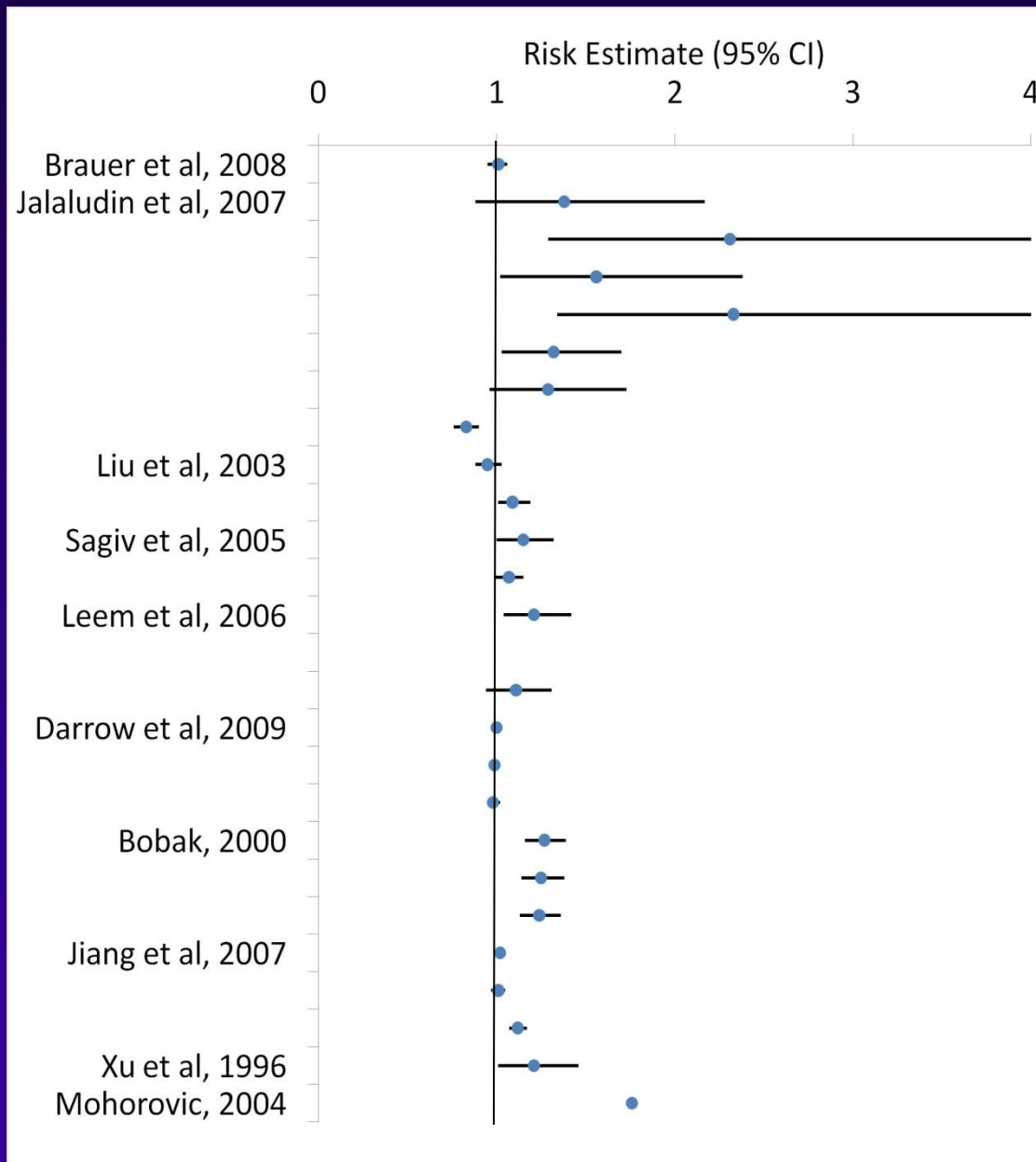
Preterm Birth

- Ten studies - eight report significant findings
- Higher SO₂ exposure - increased risk of preterm birth
- Three studies reported dose-response associations
- Studies varied
 - window of exposure
 - adjustment for distance from monitor
 - level of exposure

Reported SO₂ Exposures (ppb) in Preterm Birth Studies



Preterm Birth Forest Plot

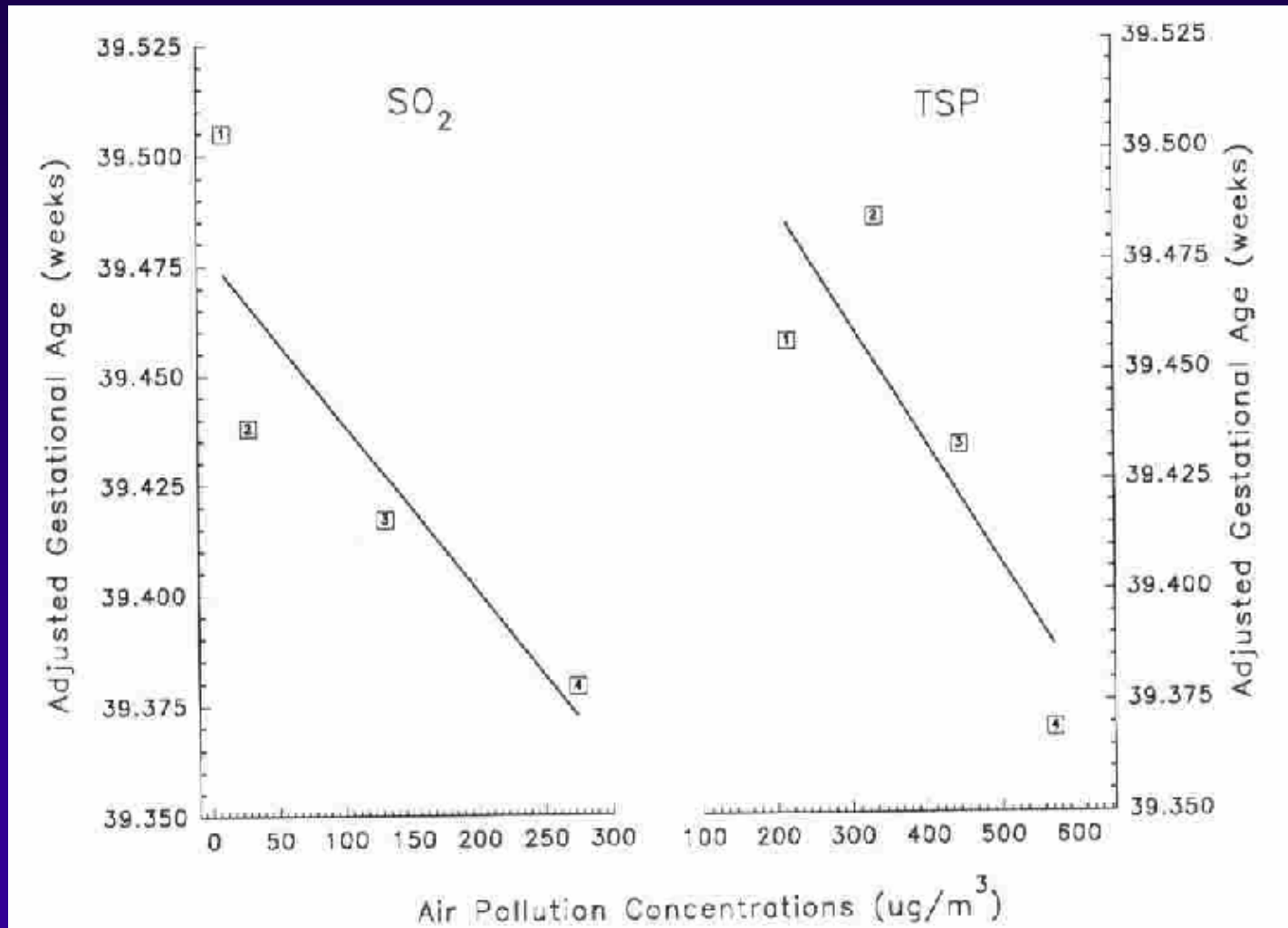


Risk estimates
with 95%
confidence
intervals from
10 studies

Study by Xu et al.

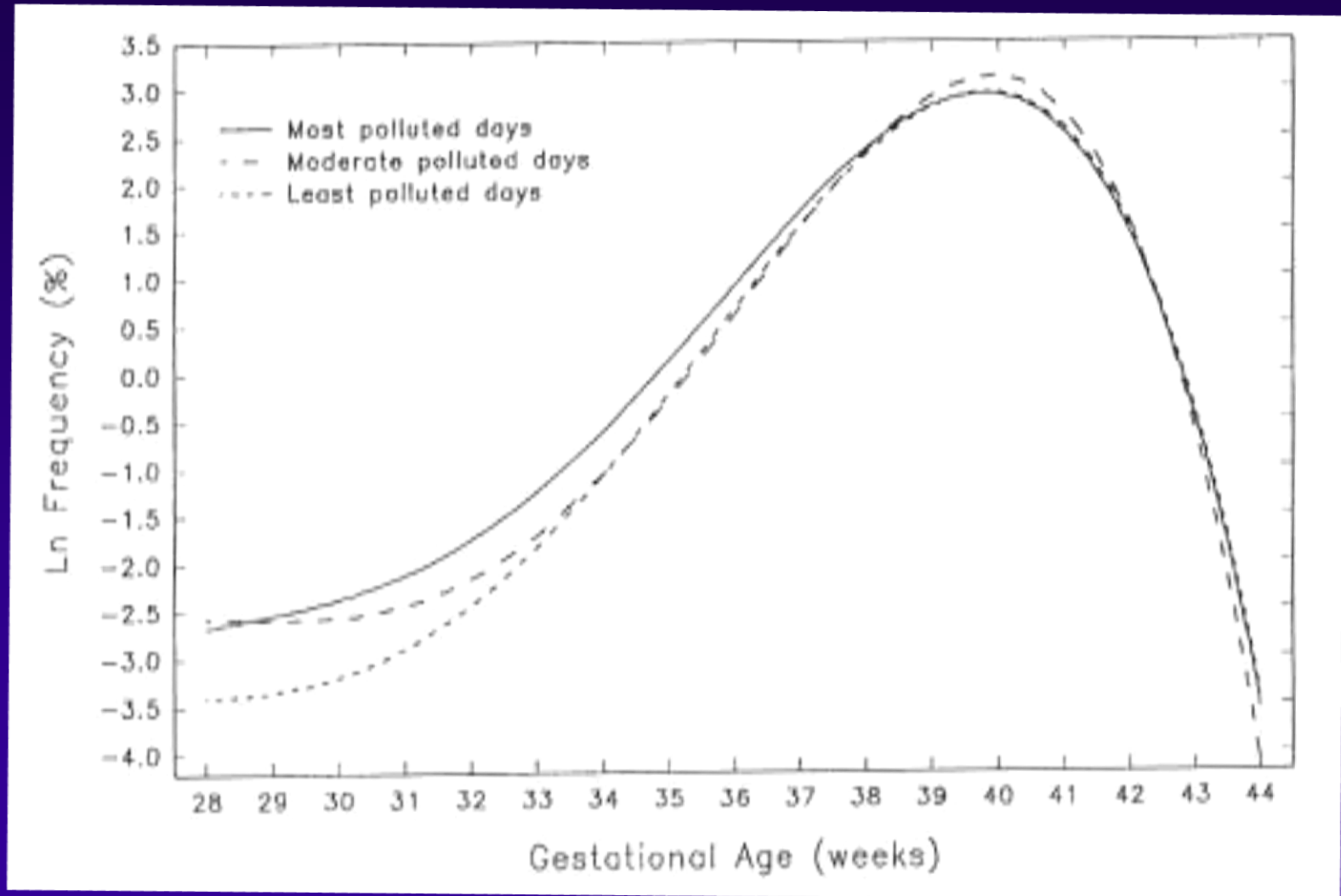
- High levels of SO₂
- A large gradient of SO₂
- Monitored and adjusted for seasonal changes
- Controlled for total suspended particles (TSP)
- Close proximity of population to air monitoring stations
- Investigated different number of lag days (exposure windows)

Adjusted Gestational Age, by SO₂ and TSP Concentrations



Derived from locally weighted regressions, adjusting for temperature, humidity, day of the week, season, maternal age, gender of child, and residential area

Gestational Age Distribution by Tertile of SO₂ Concentration



Results from the Study by Xu et al.

- Dose-dependent relationship between gestational age and SO_2 and TSP
- Adjusted odds ratio for PTB = 1.21 (95% CI= 1.01, 1.46 for each \log_e ($\mu\text{g}/\text{m}^3$) increase in SO_2
- Pregnancies at high risk for PTB may be particularly susceptible to effects of air pollution

Fetal Growth

Fetal Growth

Outcomes examined in humans

- Low birthweight (<2500 g)
- Birthweight (continuous)
- Intrauterine growth restricted (IUGR), Small for gestational age (SGA; <10th percentile for sex and gestational age)
- Very low birth weight (<1500 g)
- Fetal ultrasound scan measurements, e.g., femur length, head circumference

Fetal Growth

22 epidemiologic studies

# of Studies	Association with fetal growth
13	↑ risk only
2	↓ risk only
2	Both ↑ and ↓ risk
5	No significant associations
22	(Total)

Fetal Growth

Spatial and Temporal Exposure Assessment

Study	Association with fetal growth restriction, comments
Lin et al., 2004	↑ risk LBW at medium and high levels
Dugandzic et al., 2006	↑ risk LBW, 1 st trimester
Williams et al., 2007	Very high ↑ risk LBW; outlier
Yang et al., 2003	Slight ↓ in birth weight, 1 st trimester
Hansen, et al., 2008	↓ ultrasound measurements, low SO ₂
Brauer et al., 2008	No significant associations, low SO ₂

Fetal Growth

Co-pollutant confounding

- Carbon monoxide (CO)
- Particulate matter:
 - < 10 μm (PM₁₀)
 - < 2.5 μm (PM_{2.5})
 - Total suspended particulates (TSP)
- Nitrogen dioxide (NO₂)

Fetal Growth

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Fetal Growth

Multi-pollutant analyses

Study	Co-pollutants
Gouveia et al., 2004	CO, PM ₁₀ , (NO ₂ , O ₃)
Bobak and Leon, 1999	TSP, NO _x
Hansen et al., 2008	PM ₁₀ , NO ₂ , O ₃
Lin et al., 2004	CO, PM ₁₀ , NO ₂ , O ₃
Liu et al., 2003	CO, NO ₂ , O ₃
Williams et al., 2007	PM _{2.5} , lead
Nascimento and Moreira, 2009	PM ₁₀ , O ₃

Fetal Growth

Lin et al., 2004 - Taipei and Kaohsiung, Taiwan

- Assessed spatial and temporal variation
- Restricted cohort: 3 km from monitors
- Adjusted for CO, PM₁₀, NO₂, O₃
- Adjusted for season
- High SO₂ levels
- Exposure gradient

Fetal Growth

Lin et al., 2004

Results for LBW and SO₂ (entire pregnancy)

Exposure	SO ₂ conc. (ppb)	AOR* (95% C.I.)
Low	< 7.1	1.00
Medium	7.1 – 11.4	1.16 (1.02, 1.33)
High	>11.4	1.26 (1.04, 1.53)

* Adjusted for gestational week, gender, birth order, season, maternal age & education, and co-pollutants (CO, PM₁₀, NO₂, O₃)

Fetal Growth

Lin et al., 2004

Results for LBW and SO₂ (third trimester)

Exposure	SO ₂ conc. (ppb)	AOR* (95% C.I.)
Low	< 6.8	1.00
Medium	6.8 – 12.4	1.13 (0.99, 1.28)
High	>12.4	1.20 (1.01, 1.41)

* Adjusted for gestational week, gender, birth order, season, maternal age & education, and co-pollutants (CO, PM₁₀, NO₂, O₃)

Fetal Growth

Lin et al., 2004

Limitations

- Kaohsiung and Taipei
- Maternal characteristics
- CO was associated with ↓ risk of LBW
- Correlations among pollutants not reported

Fetal Growth

Animal studies

- ↓ birthweight in mice at 65,000 ppb; concentration-dependent
- ↓ fetal weight, no change in crown-rump length in mice at 25,000 ppb
- No effect on fetal weight in rabbits at 70,000 ppb

Congenital Malformations

Congenital Malformations

Epidemiologic Studies

Methodological challenges

- Confounding
- Multiple comparisons
- Case identification
- Case groupings, syndromes

Congenital Malformations

Epidemiologic Studies

Case groupings

- Any/all birth defects
- Chromosomal vs. non-chromosomal defects
- Heart defects
- Oral clefts

Inconsistent findings

Congenital Malformations

Animal study

- No association with specific or aggregate malformations in mice at 25,000 ppb or rabbits at 70,000 ppb SO₂

Pregnancy Loss

Pregnancy Loss

Spontaneous abortion

- No association in a cross-sectional occupational study

Pregnancy Loss

Stillbirth

- Fetal death after 28 weeks gestation, or $>1,000$ g
- Ecologic studies
- Correlation with SO_2 ($r=0.7$; $p \leq 0.05$)

Pregnancy Loss

Animal studies

Gestational exposure to SO_2 did not result in changes in mean litter size or resorption frequencies at:

- 25,000 ppb (mice)
- 70,000 ppb (rabbits)

Exposure to 32,000 ppb or 65,000 ppb SO_2 was not associated with changes in litter size

Asthma

Prenatal exposure to SO₂ associated with ↑ risk of childhood asthma...

- High correlations between pre- and postnatal exposure
- High correlations among co-pollutants
- Traffic-related pollutants had stronger associations

Other Developmental Outcomes

Mice

- Effects on male-male social behavior at 12,000 & 30,000 ppb
- Delays in acquisition of certain postnatal reflexes at 32,000 ppb

Developmental Toxicity

Summary

- Preterm birth
- Low birth weight
- Congenital malformations
- Pregnancy loss
- Asthma
- Other developmental effects

Questions?

Female Reproductive Toxicity

Female Reproductive Toxicity

In vitro fertilization (IVF)

- SO₂ was consistently but not significantly associated with ↓ odds of live birth
- Other pollutants more strongly associated with odds of live birth

Female Reproductive Toxicity

Animal Study

Rats exposed by inhalation to SO₂ at ~1,500 ppb showed effects on:

- Estrous cycle length in F₀ and F₁ offspring
- Pregnancy frequency and duration
- Offspring growth (body weight)
- No changes observed at ~57 ppb

Female Reproductive Toxicity

Related studies: Sodium sulfite

- *In vitro* exposure of sheep or cow oocytes resulted in fragmentation of chromosomes, with or without rearrangement
- No effects were seen in mouse oocytes exposed either *in vitro* or *in vivo*

Female Reproductive Toxicity Summary

- Humans: IVF
- Animals: Estrous cycles and pregnancy
- Related studies

Questions?